

REMARKS

3. The Examiner pointed out that claim 17 is dependent on a cancelled claim. The claim has now been corrected by making it dependent on claim 53.

Claim Rejections

4. The Examiner has provisionally rejected claims 53 and 57 under obviousness-type double patenting as being unpatentable over the claims of allowed application No. 09/330,594 (which issued on September 3, 2002 as US patent No. 6,444,467). Claim 57 has now been cancelled, so the provisional rejection of this claim is now moot. As for the rejection of claim 53, reconsideration of this rejection is again requested. Claim 53 has now been amended to require the controlled environment to provide a relative humidity of 90 to 100% for at least 2 days after sowing. This is a feature that is not present in the claims of 09/330,594 and is not obvious therefrom.
5. The Examiner maintained the provisional rejection of claim 45 under obviousness-type double patenting as being unpatentable over claims 1-35 of Application no. 09/550,110. Applicant requests reconsideration of this rejection for the following reasons. Claim 45 includes the limitations of amended claim 53. As noted above, claim 53 has now been amended to require the controlled environment to provide a relative humidity of 90 to 100% for at least 2 days after sowing. This is a feature that is not present in the claims of 09/550,110 and is not obvious therefrom.
6. 7. & 8
- The Examiner rejected claims 5, 10-11, 15-20, 24-28, 30, 45 and 53-57 as anticipated by Dupuis et al. To overcome this rejection, independent claim 57 has been cancelled and independent claim 53 has been amended.

The amendments made to claim 53 address the issues set out by the Examiner in the final rejection. That is to say, claim 53 now requires the three-phase substrate to be a growth medium

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selected from soil, peat and horticultural growing mixes, which is clearly intended from the specification as originally filed. In fact, since the claim has been limited in this way, all reference to the use of a three-phase substrate has been removed from the claim. It will be apparent that soil, peat and horticultural growing mixes prepared for sowing have the required three phases required of the original claims. The term "placing" has also been changed to "*ex vitro* sowing" for greater clarity and to distinguish more clearly from *in vitro* germination techniques, and there is a requirement that the embryos be "unencapsulated" when sown. This term clear in meaning and avoids the Examiner's criticism of the word "naked". The term has been used in the specification as filed, and is therefore supported by the disclosure. It is believed that these amendments clearly show that the present invention does not apply to germination in a gel or capsule of the kind disclosed in the prior art, such as Dupuis et al and Carlson et al. The present invention is concerned with the germination of somatic embryos in non-sterile soil, peat or common growing mixes that make the procedure substantially equivalent to the planting of conventional zygotic seeds in non-sterile conditions.

Additionally, amended claim 53 now requires the application of a nutrient solution by way of fogging, misting or irrigation.

Importantly, amended claim 53 also requires the provision of a relative humidity in the environment of at least 90% for at least 2 days after sowing. This limitation is supported by the disclosure (e.g. page 14, lines 1 to 3). This ensures viability of the embryos in non-sterile conditions during a critical phase of their development. Such high relative humidities are outside the ranges employed in conventional sowing techniques.

In addition to this, the claims of this invention have now been restricted to gymnosperm species.

The present invention addresses the problem of converting somatic embryos into seedlings and plants in a manner that can be operated on a commercial level at an economic cost, and preferably using conventional seeding equipment. As noted in the introduction of the present application, many approaches to this problem have involved either encapsulation of the embryos or alternatively the conversion of naked (unencapsulated) embryos in sterile conditions. None of these approaches has been successful on a commercial basis, at least for gymnosperm species. In contrast, Applicants found that somatic embryos can be sown naked (unencapsulated) directly



into or onto commercial-type non-sterile growing media with good conversion rates, provided that certain steps are carried out. In particular, a very high level of humidity is required for a minimum period immediately after sowing. Secondly, a carbohydrate nutrient (e.g. a sugar) must be present during embryo germination.

Dupuis et al. is concerned with the conversion of embryos primarily in a gel medium and the claims of the present invention have been distinguished more clearly from this approach by restricting the claims to sowing directly into non-sterile soil, peat or a horticultural growing mix. The basis of Dupuis et al. is to allow germination in a non-sterile environment (i.e. an environment open to the atmosphere) basically by eliminating all pathogenic organisms from the gel medium by adding a fungicide or bactericide to the medium. In essence, therefore, Dupuis et al. teaches that germination in a non-sterile environment is only acceptable if the growing medium (gel substrate) is made aseptic by application of effective amounts of a fungicide and bactericide. While the present application allows for the use of such germicides, they are not essential and Applicants have found a way to carry out germination even in the absence of such materials even when sowing the embryos into a non-sterile growing medium such as conventional soil or horticultural mix. The present invention allows for the use of such fungicidal or bactericidal materials. The fact remains, however, that the present invention can be carried out successfully without resorting to the use of fungicides or bactericides, which makes it distinct from Dupuis et al.

As noted above, the present invention requires the use of high humidity during the initial stage of germination after sowing. The humidity must be kept above 90% during this phase, which is a very high level. Dupuis et al. mentions a humidity level of only 30 to 70% (page 6, line 5 of the English translation) and thus does not disclose or suggest the present invention. There is no reason why a person skilled in the art, after reading Dupuis et al., would consider raising the relative humidity significantly above the 70% maximum of the indicated range. Therefore this feature of the present invention is not obvious from Dupuis et al.

Finally, the Examples in Dupuis et al. relate only to carrot - an angiosperm specie. The claims of the present invention are now restricted to gymnosperm. While Dupuis et al. mentions various species, including gymnosperms, on page 2, paragraph 3 of the translation, there is no experimental verification of this.

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In summary, it is Applicant's view that Dupuis et al. discloses an invention that is different from the present invention, as now claimed, and there is no disclosure or teaching of that invention anywhere in Dupuis et al.

Carlson et al. (US 5,486,218) is not seen as relevant to the present invention at all. This reference relates to the encapsulation of embryos, rather than the sowing of such embryos in a naked or unencapsulated condition. The invention is concerned primarily with the provision of oxygen for the embryo when it is encapsulated, as the capsule can limit access to oxygen. Obviously, this addresses a quite different situation than that of the present invention where the sowing of unencapsulated embryos causes no restriction to the access of oxygen.

Fuji et al. relates only to the sowing of embryos of alfalfa - an angiosperm. The paper is primarily concerned with the effect of ABA or mannitol during embryo maturation rather than the effects of treatments during germination.

Thus, the prior art, taken alone or in combination, does not disclose the combination of feature of the present invention, namely the application to gymnosperm species, the sowing into non-sterile growing media, the use of high relative humidities during initial germination and the presence of carbohydrate result in a novel and unobvious set of conditions that enable conversion of somatic gymnosperm somatic embryos in a manner substantially equivalent to zygotic seed.

In view of the above amendments and comments, favourable reconsideration of this application is requested.

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Respectfully submitted,



Marked-up copy of the amended claims showing changes made

5.(Amended) The process of claim 53 wherein [said] at least one environmental factor of said controlled environment [is] selected from the group consisting of a moisture level within said three-phase substrate, [atmospheric humidity,] temperature, nutrients, ambient light intensity and diurnal photoperiod is manipulated during embryo germination to facilitate germination of the somatic embryo.

Cancel claims 8 and 9.

Cancel claim 15 and 16.

17.(Amended) The process according to claim [1] 53 wherein the somatic embryo is a somatic embryo which has been previously desiccated to a final moisture content in the range of 5 – 75%.

Cancel claim 18

19.(Amended) The process according to claim 53 wherein the [solid phase of the three-phase substrate] growing mix is a mixture of substrates selected from the group consisting of peat, sawdust, bark chips, wood chips, compost, moss, perlite, vermiculite, pumice, grit, sand, soil, cellulosic fibres of plant origin, extruded foams, extruded fibres, and chemically expanded foams.

20.(Amended) The process according to claim 53, wherein the [three-phase substrate] growth medium contains a wetting agent.

22.(Amended) The process according to claim 53 wherein the moisture content of the [three-phase substrate] growth medium is adjusted with water to a range of 60-85% prior to receiving a somatic embryo.

23.(Amended) The process according to claim 53 wherein the moisture content of the [three-phase substrate] growth medium is adjusted with a nutrient solution to a range of 60-85% prior to receiving a somatic embryo.

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24.(Amended) The process according to claim 53 wherein at least one fungicide to control plant pathogens is incorporated into the [three-phase] growth [substrate] medium.

25.(Amended) The process according to claim 53 wherein at least one fungicide to control plant pathogens is applied in liquid form to the [three-phase substrate] growth medium.

26.(Amended) The process according to claim 53 wherein at least one fungicide to control plant pathogens is applied in aerosol form to the [three-phase substrate] growth medium.

27.(Amended) The process according to claim 53 wherein at least one insecticide to control plant pests is incorporated into the [three-phase growth substrate] growth medium.

28.(Amended) The process according to claim 53 wherein at least one insecticide to control plant pests is applied in liquid form to the [three-phase substrate] growth medium.

29.(Amended) The process according to claim 53 wherein at least one insecticide to control plant pests is applied in aerosol form to the [three-phase substrate] growth medium.

30.(Amended) The process according to claim 53 wherein the [three-phase substrate] growth medium is contained within a horticultural container.

34.(Amended) The process according to claim 53 wherein the somatic embryo placed on or within the [three-phase substrate] growth medium, is covered with a material selected from the group consisting of peat, sawdust, bark chips, wood chips, compost, moss, perlite, vermiculite, pumice, grit, sand, soil, cellulose fibres of plant origin, extruded foams, extruded fibres, and chemically expanded foams.

35.(Amended) The process according to claim 53 wherein the somatic embryo is placed on or within the [three-phase substrate] growth medium with seeding equipment.

Cancel claim 39 and 40.

41.(Amended) The process according to claim [39] 53 wherein the carbohydrate is a [sugars are] sugar selected from the group consisting of monosaccharides and polysaccharides.

42.(Amended) The process according to claim [39] 53 wherein the carbohydrate is a [sugars are] sugar selected from the group consisting of glucose, fructose, mannose, maltose, and sucrose.

43.(Amended) The process according to claim 53 wherein only water is applied [as microdroplets] to the surface of the [three-phase substrate] growth medium for a period of 18-36 hours after [placing] sowing the somatic embryo on or within the [surface of the three-phase substrate] growth medium, after which time, nutrient solutions are also applied [as microdroplets].

45.(Amended) A process of growing a somatic embryo into a seedling, which comprises maintaining a somatic embryo germinated according to the process of claim 1 in a [three-phase substrate] growth medium, and growing said germinated embryo to develop the germinated embryo into a seedling.

53.(Amended) A process of germinating gymnosperm somatic embryos, which comprises:

- (a) [placing] ex vitro sowing [a] an unencapsulated gymnosperm somatic plant embryo on or within a non-sterile [three-phase substrate] plant growth medium[, the phases comprising solid, liquid and gas phases,] selected from the group consisting of peat, soil and a horticultural growing mix.
- (b) placing the [substrate] medium containing the somatic embryo into [an] a non-sterile environmentally-controlled plant-growing environment in which at least [one environmental factor] relative humidity may be controlled and manipulated,
- (c) [manipulating at least one factor] maintaining said relative humidity in the range of 90 to 100% for at least two days from sowing to enable and facilitate germination of the somatic embryo, and
- (d) making a nutrient carbohydrate available to the embryo, and applying nutrient [solutions] solution by a method selected from the group consisting of fogging, misting and irrigation, [at regular intervals] at least during the period of somatic embryo germination

[for a period of time] such that somatic embryo imbibition, germination, growth and development occur.

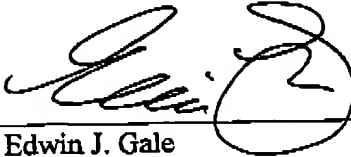
Cancel claims 54, 55 and 57

56.(Amended) A process according to claim 53 wherein the [water and/]nutrient solution is applied in the form of microdroplets.

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